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EFFECTS OF NITROGEN GAS FUMIGATION ON POSTHARVEST QUALITY OF MINIMALLY PROCESSED STARFRUIT (*Averrhoa carambola* L.) STORED AT LOW TEMPERATURE

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ABSTRACT

Minimally processed (MP) fruit have a short post-cutting life due to the mechanical injury during processing which increase the browning incidence, tissue softening, as well as the nutrient loss and flavor. The browning incidence occurs due to the destructive of cellular compartmentation which allow the phenolic substrate to be assessable to Polyphenol Oxidase (PPO) that catalyze phenolic oxidation. Therefore, the appearance, organoleptical properties and nutritional quality are reduced in MP starfruit. In this study, the effect of six different volumes of nitrogen gas (N₂) (0 mL, 5 mL, 10 mL, 15 mL, 20 mL and 25 mL) fumigation on postharvest quality of MP starfruit (*Averrhoa carambola* L.) stored at 5°C were investigated. Flesh colour, browning index, total phenolic content, soluble solids concentration (SSC), titratable acidity (TA), firmness and ascorbic acid concentration of MP starfruit were determined for 12 days period. No significant differences of various N₂ fumigation treatments were recorded in all postharvest parameters except for percentage weight loss. However, N₂ fumigation at higher volume, 25 mL was a promising value to reduce browning index as well as maintain MP fresh colour and total phenolic content. In conclusion, the application of 25ml N₂ gas has a tendency to be the best volume in reducing browning incidence and maintaining other quality attributes of MP starfruit.

Key words: fresh cut, enzyme, fumigation, postharvest quality

INTRODUCTION

Starfruit (*Averrhoa carambola* L.) is a popular tropical fruit found in Southeast Asia and is classified under Oxalidaceae family. In Malaysia, two clones of starfruit, namely B10 and B17, has attained a commercial status (Abd Rahman *et al.*, 2007) and widely grown in the states of Johore, Selangor and Negeri Sembilan. For clone B10, starfruit originated from Serdang Baru, Selangor 1968, it is oblong, sweet (8-10°Brix), aromatic, juicy, gold yellow colour and fine texture. Meanwhile, clone 17 originated from Triang, Pahang 1988, it is cylinder shape, large, fibrous, juicy, crispy, aromatic, red golden colour and very sweet (12-18°Brix) (Department of Agricultural Malaysia, 2010). Starfruit has been reported to have various therapeutic effects such as relieve bleeding haemorrhoids, haemorrhages, fevers, biliousness, diarrhoea and to relieve a “hangover” from

excessive indulgence in alcohol (Morton, 1987). Due to its unique flavor and texture, high in nutritional and bioactive compounds, starfruits are categorized as the most highly consumed and an extremely important agricultural trade product in Malaysia. However, starfruit is highly perishable and easily deteriorate due to its respiration behaviour during ripening process. Besides, its ripening process was accelerated when stored at high temperature due to the high respiration rate. In addition, the browning of the rib-edge due to the physical injury during marketing and storage also contributed to the low quality of starfruit. This browning incidence can intensify with water loss. Fruit that have lost about 5% of their weight due to water loss show visible symptoms of dehydration.

Browning has attributed to the action of polyphenoloxidase (PPO) on the natural phenolic substances or compounds of the fruits (Nicoli *et al.*, 1994). The browning of the fresh cut starfruit is due to the destruction of fruit cellular compartmentation which allows the phenolic substances to be

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assessable to PPO that catalyze the phenolic oxidation (Ding *et al.*, 2007). Hence, the appearance of fresh cut starfruit is therefore destructive and the organoleptical properties and nutritional quality are reduced. Thus, further investigation for prolonging the shelf life of the fresh cut starfruit is highly needed.

Recently, fresh starfruit that is cut transversely into star-shaped can serve as an addition to salads to form new product to meet the needs of consumers for convenience. However, minimally processed (MP) fruits have a short post-cutting shelf-life as discussed above. Many researches have been conducted to reduce the browning of the fresh cut fruit such as in starfruit (Ding *et al.*, 2007), mango (Gonza'lez-Aguilar *et al.*, 2000), rose apple (Suriyan *et al.*, 2012) and apple (Lee *et al.*, 2003). However, there is lack of information available on the effect of nitrogen (N_2) gas fumigation and low temperature storage in reducing browning incidence of MP starfruit without significant reduction of its quality. Therefore, this study aimed to evaluate the effect of different volumes N_2 gas fumigation in delaying browning incidence and maintaining the quality of MP starfruit stored at low temperature and also to determine the best volume of N_2 gas to be applied. By looking on the importance of the fresh cut starfruit in domestic as well as the international markets, there is an urgent need to explore a biologically safe, low cost treatment to reduce browning and maintain the postharvest quality of minimally processed starfruit.

MATERIALS AND METHODS

The experiment was conducted at the Postharvest Technology Laboratory, School of Food Science and Technology, Universiti Malaysia Terengganu. Starfruits at maturity stage 4 were freshly purchased from commercial orchard at Fruit Valley, Selangor. A total of 216 starfruits with similar size, maturity, free from defects and decay were chosen. The starfruits were washed and soaked by using 0.02% (200 ppm) sodium hypochloride solution for 10 minutes. Then, each starfruit was cut in five slices with approximately 1 cm thick for each slice. The knife and cutting board were sanitized with 200 ppm sodium hypochloride solution for 3 minutes prior to use. The starfruit slice fruits were sorted and arranged as according to the assigned treatments. Eighteen of air-tight polyvinyl chloride (PVC) containers (1080 mL) were used in the experiment. Each container contain 30 slices of starfruits and arranged according to Completely Randomized Design (CRD) with three replications. The sealed PVC containers which then injected with the desired volumes of N_2 through an injection port. The N_2 gas

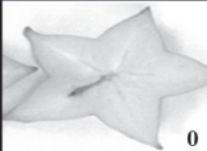


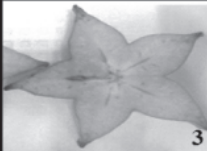
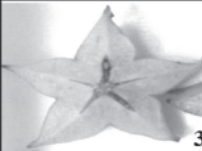
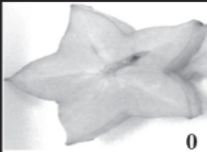
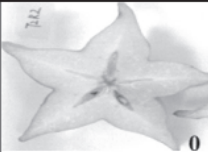

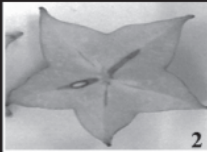


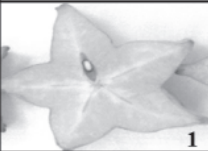

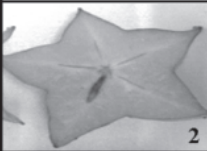



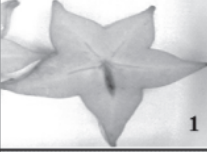


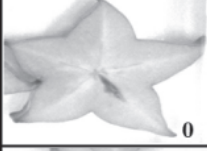
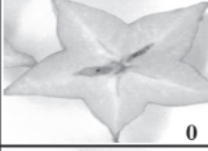

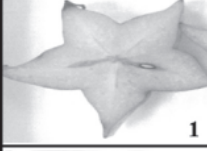
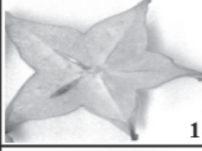



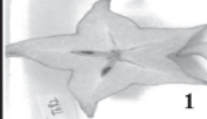

was obtained from MCY Company Gas. Six different volumes of N_2 gas were fumigated on starfruit slice fruits viz. (i) control (0mL), (ii) 5 mL, (iii) 10 mL, (iv) 15 mL, (v) 20 mL, (vi) 25 mL and kept at 5°C for 20 mins, RH 90-95% in the PVC containers. After 20 mins, five slices of starfruit were placed on polypropylene trays and then was sealed with polyethylene film.

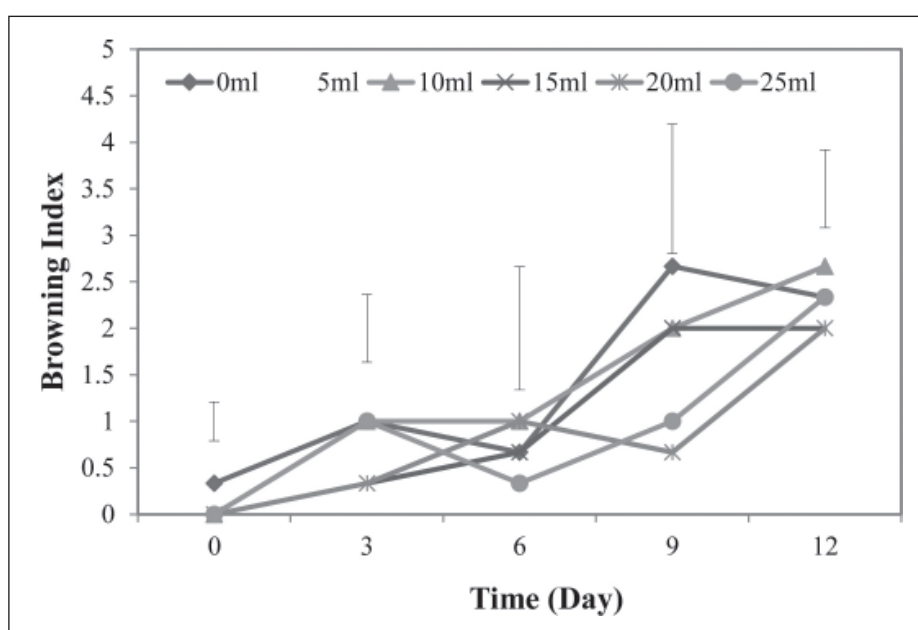
Parameters evaluated were fruit colour, browning index, soluble solids concentration, flesh firmness, total phenolic content and ascorbic acid concentration. Colour of starfruit flesh was assessed by using a Minolta Chroma Meter (Model R200 Trimulus Colour Analyser, Minolta camera Co. Ltd., Japan). Colour data were expressed in L^* , a^* and b^* values. L^* represented the lightness coefficient which ranges from 0 (black) to 100 (white). a^* ranged from -60 to +60, which indicates red (+60) and green (-60) colours. Meanwhile, b^* ranged from -60 to +60, which indicates as yellow (+60) and blue (-60) colours. a^* and b^* were further used to calculate hue angle ($h^\circ = \tan^{-1} b^*/a^*$) for colour interpretation. Hue angle (h°) represented red-purple (0°), yellow (90°), bluish green (180°) and blue (270°) (McGuire, 1992). Meanwhile, browning index was estimated by measuring the extent of the total brown area on each fruit surface using the following scale: 0 = none, 2 = trace, 3 = slight, 4 = severe and, 5 = extremely severe (Ding *et al.*, 2007). Total phenolic content was evaluated by using a modified colorimetric method as described by Singleton and Rossi (1965). Meanwhile, ascorbic acid concentration was based on the method of AOAC (1984). The parameters were assessed on every three-day interval up to twelve days viz. day 0, 3, 6, 9 and 12. The data were subjected to the analysis of variance (ANOVA) using GLM (General Linear Models) procedures with SAS 9.3 software package, SAS Institute Inc, Cary, NC, USA. Treatments means were further separated by LSD for least significance at $P \leq 0.05$ (SAS Institute Inc., 1999).

RESULTS AND DISCUSSION

Visual quality is one of the important factors in determining the market value of fresh fruits and vegetables. The browning of MP starfruit began from fruit margin then progressed to the cut surface (Ding *et al.*, 2007). In the present study, MP starfruit fumigated with the N_2 gas had similar effect with control fruit in reducing browning incidence. As shown in Table 1 and Figure 1, no apparent effect of different volumes of N_2 gas was observed on browning incidence of MP starfruit. All gas treatments had similar effects on browning index, however, on day 9 and day 12 MP starfruits treated

Table 1. The browning index on MP starfruit treated with different volume of nitrogen gas fumigation. Number in the box denotes to browning score of MP starfruit.

Treatment Time	Day 0	Day 3	Day 6	Day 9	Day 12
0mL of N ₂ gas	 0	 1	 3	 3	 3
5mL of N ₂ gas	 0	 0	 1	 2	 3
10mL of N ₂ gas	 0	 1	 1	 2	 3
15mL of N ₂ gas	 0	 0	 1	 2	 2
20mL of N ₂ gas	 0	 0	 1	 1	 1
25mL of N ₂ gas	 0	 0	 0	 1	 2

**Fig. 1.** Effect of different volumes of nitrogen gas fumigation on browning index of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=0.42, Day 3=0.73, Day 6=1.33, Day 9=1.39 and Day12=0.84).

with 20 mL and 25 mL of N_2 gas tend to have the lower score of browning index with the score of 0.67 and 2 respectively. N_2 gas fumigation at 25 mL had score 1 on day 9 while score 3 on day 12 (Table 1). Besides, the browning index on MP starfruit treated with different gas treatments were increased throughout 12 days experimental period. However, the browning incidence on MP starfruits fumigated with higher volumes, 20 mL and 25 mL of N_2 gas seem to be more effective to reduce the browning of the cut surface of starfruits compared with other treatments throughout 12 days storage. In general, the flesh colour of MP starfruit becomes progressively browner and less pure yellow than currently cut fresh starfruits. This coincides with the browning score of MP starfruit that visually assessed in which 20 mL and 25 mL of N_2 gas resulted in the lowest browning incidence as compared to other treatments. Possibly, the lower browning index might be due to the higher residual N_2 gas in micropores on starfruit tissue that can suppressed the enzyme activity as reported by Wu *et al* (2012a). In addition, Zhan and Zhang (2005) claimed that gas hydrates in starfruit tissue may reduce the water activity in fruit tissue and influence the protein structure of enzyme and hence the enzyme activity is restrained thereby reduce the incidence of flesh browning.

The browning incidence of MP starfruit also can be explicated based on the flesh colour. The colour parameters evaluated include lightness (L^*), chromaticity a^* , b^* , Chroma (C^*) and hue angle

(h°). The lower values of L^* , b^* , h° and higher a^* and C^* indicates browning of the fruit. The L^* , a^* and h° of MP carambola did not significantly affect ($P \geq 0.05$) with different volumes of N_2 gas except day 12 (Figure 2, 3 and 4). Meanwhile, chromaticity b^* and C^* of MP carambola were not affected ($P \geq 0.05$) with different volumes of N_2 gas fumigation throughout 12 days experimental period (Figure 5 and 6). Regardless of N_2 gas treatments, the values of L^* , a^* and h° of MP carambola were fluctuated throughout 12 days experimental period. The L^* value of MP carambola were ranging between 49.00 L^* and 55.00 L^* while a^* value were ranging from 0.60 a^* to 2.10 a^* . The h° value of MP carambola were recorded at the ranged between 78.00 h° and 87.00 h° . The higher value of L^* , chromaticity b^* , h° and lower value of chromaticity a^* and C^* denotes to low browning flesh. MP starfruits fumigated with 25 mL of N_2 gas tend to had higher L^* and lower a^* and C^* of values as compared to other treatments which reflected to the lower browning incidence (Figure 2, 3, 4, 5, 6). Similarly, high pressure nitrogen gas treatments were efficient in eliminating the browning incidence on pineapple wedge surfaces at 4°C as reported by Wu *et al* (2012a).

Phenolic compounds are very important fruit constituents due to their antioxidant activity in chelating redox-active metal ions, inactivating lipid free radical chains and preventing hydroperoxide conversion into reactive oxyradicals (Oliveira *et al.*, 2009). The involvement of phenylalanine (PAL),

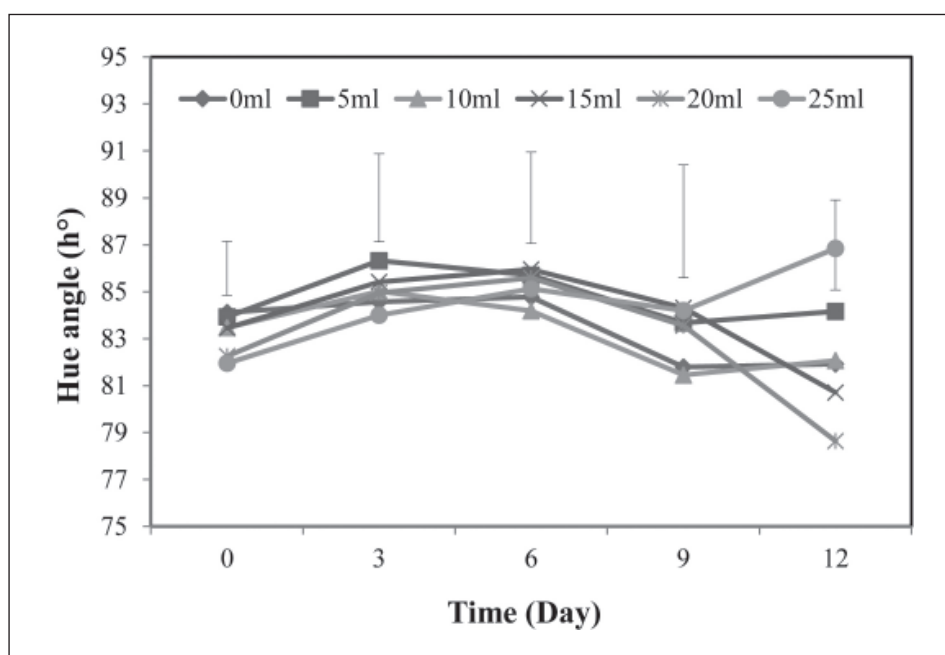


Fig. 2. Effect of different volumes of nitrogen gas fumigation on lightness of MP starfruit flesh. Vertical bars represent LSD at $P \leq 0.05$. (LSD value: Day 0=2.37, Day 3=3.13, Day 6=2.85, Day 9=2.80 and Day12=2.00)

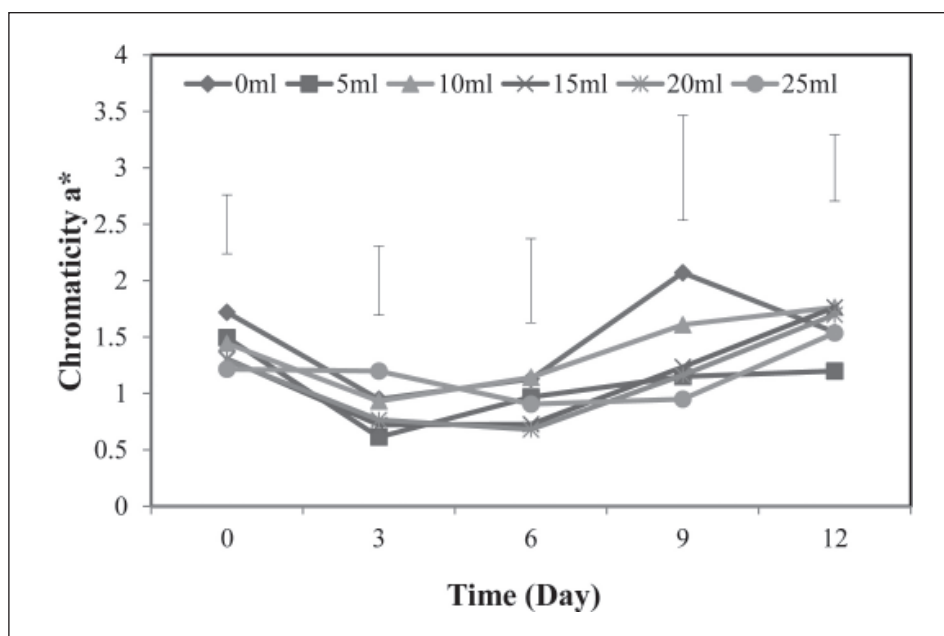


Fig. 3. Effect of different volumes of nitrogen gas fumigation on hue angle of MP starfruit flesh. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=2.30, Day 3=3.76, Day 6=3.91, Day 9=4.81 and Day12=3.83)

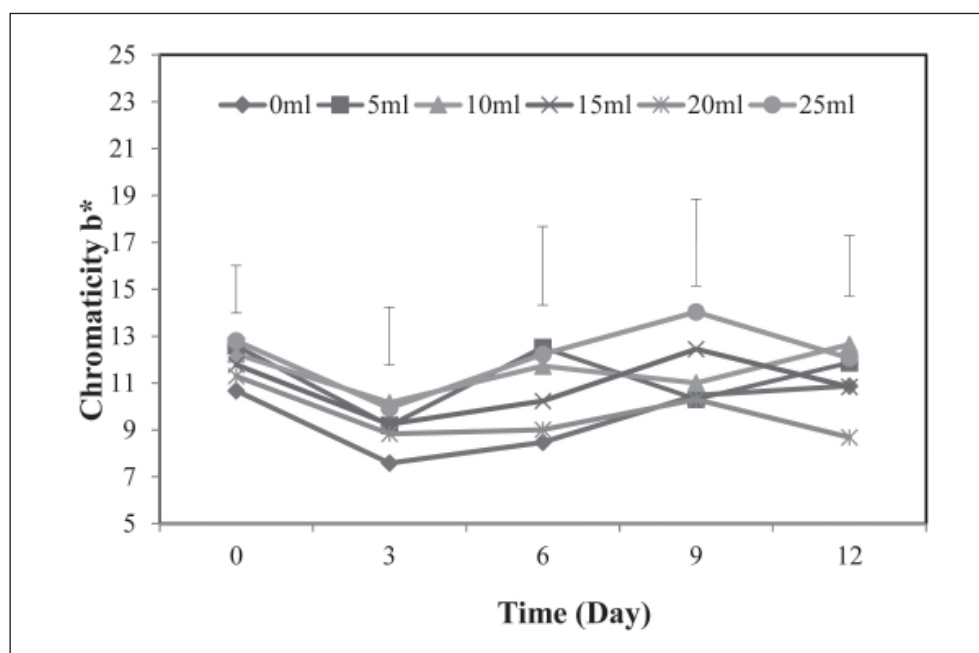


Fig. 4. Effect of different volumes of nitrogen gas fumigation on chromaticity b^* of MP starfruit flesh. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=0.52, Day 3=0.62, Day 6=0.74, Day 9=0.93 and Day12=0.59)

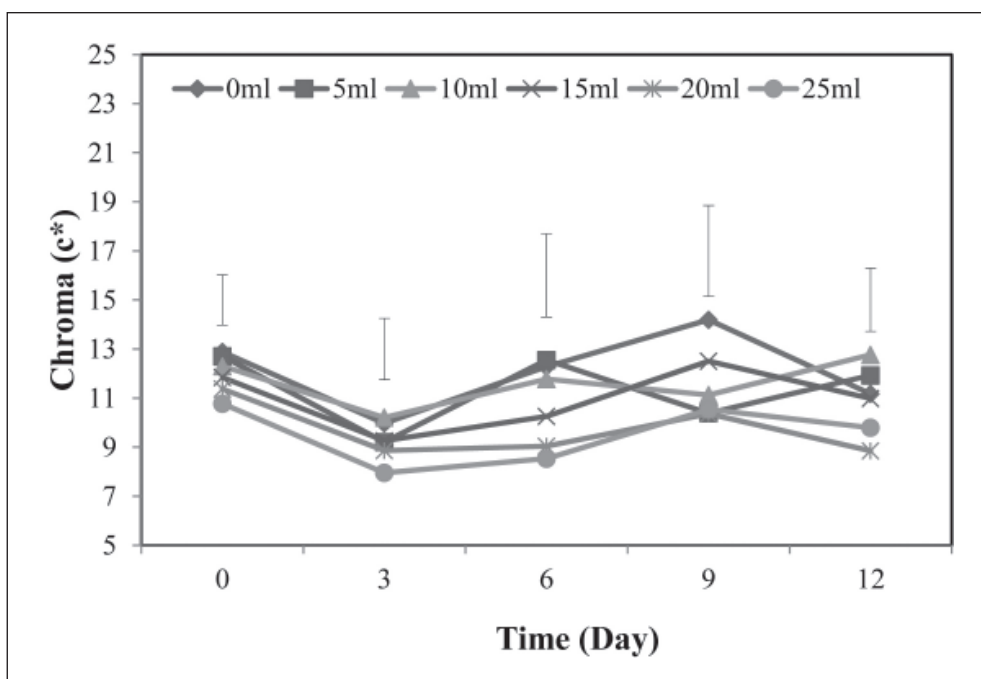


Fig. 5. Effect of different volumes of nitrogen gas fumigation on chromaticity value b of MP starfruit flesh. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=2.03, Day 3=2.49, Day 6=3.36, Day 9=3.70 and Day 12=2.59).

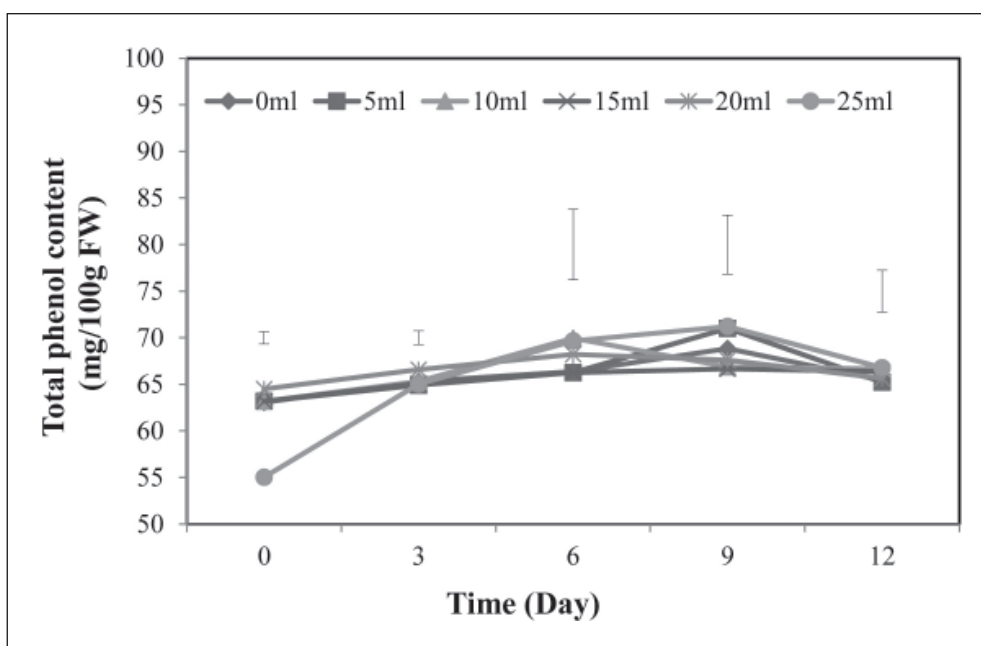


Fig. 6. Effect of different volumes of nitrogen gas fumigation on chroma of MP starfruit flesh. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=2.03, Day 3=2.50, Day 6=3.38, Day 9=3.71 and Day 12=2.57).

polyphenol oxidase (PPO), and peroxidase (POD) play important role in enzymatic browning of many fruits and vegetables. PAL is a key enzyme of phenolic synthesis and acts on the conversion of L-phenylalanine to trans-cinnamic acid in the phenylpropanoid pathway (Zhu *et al.*, 2009). Similarly, Siddiqui *et al* (2011) claimed that PAL is a key enzyme in synthesizing the phenolic compounds which oxidized by PPO resulted in tissue browning. Walker (1995) also reported that PPO and POD can be attributed to the oxidation of phenolic compounds to form brown pigment. In the present study, there are no visible effects of different volumes of N₂ gas on total phenolic content of MP starfruit throughout 12 experimental period (Figure 7). The browning incidence was reduced might be attributed to the residual of nitrogen gas in microspores of starfruit tissues that inhibited the PAL, PPO, and POD activities and decreased the browning incidence same as found on MP pineapple at 4°C after high pressure nitrogen treatment applied which reported by Wu *et al* (2012a). In addition, the reduction of browning incidence might be attributed to the formation of gas hydrates in fruit tissue and manipulated the protein structure of enzyme restrained the incidence as claimed by Wu *et al* (2012a).

On the other hand, no apparent trend on ascorbic acid (AA) concentration of MP starfruit in all treatments throughout 12 days experiment period was observed (Figure 8). However, the AA concentration of MP starfruits fumigated with N₂ gas did not have much difference with the control fruit. In general, the AA concentration of MP starfruits

ranged from 28mg/100g FW to 34mg/100g FW. United State Department of Agricultural (2014) has reported that fresh starfruit generally contains total ascorbic acid with 34.4mg/100g FW. This proved that no adverse effect of N₂ gas fumigation on the AA content in the MP starfruit. This finding was similar with the results of Wu *et al* (2012a) who reported that a moderate reduction in pineapple wedges treated with N₂ gas at 4°C. However, further investigation is required to explain the role of N₂ gas in maintaining AA content.

The soluble solid concentration (SSC) of MP starfruits treated with different volumes of N₂ gas treatment were maintained as no pronounced effect were observed as shown on Figure 9. Wu *et al* (2012b) also reported that no adverse effect of inert argon gas for 14 days on SSC of apple slices. The increased value in sugars of MP starfruit might be attributed to the ripening process. Meanwhile, the decreased in sugars in MP starfruit might be due to the oxidation of substrate that occurred during respiration process in high oxygen content. Meanwhile, the titratable acidity (TA) of MP starfruit treated with different volumes of N₂ gas had increased and decreased inconsistently from day 0 to day 12 in all treatments (Figure 10). No specific trend can be deduced for SSC and TA of MP starfruit. The reason of increase and decrease value of SSC and TA throughout the experimental period was unknown. Further investigations are needed to explain these phenomena. The fluctuation of SSC and TA in MP starfruit was similar to the report of Ding *et al* (2007).

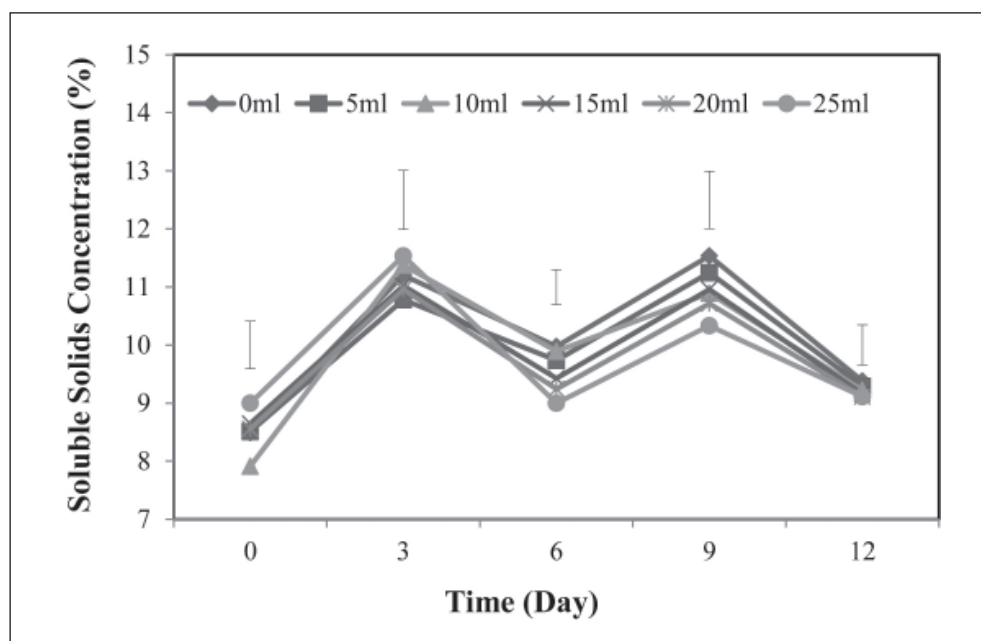


Fig. 7. Effect of different volumes of nitrogen gas fumigation on total phenolic content of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=1.32, Day 3=1.46, Day 6=7.55, Day 9=6.31 and Day12=4.47)

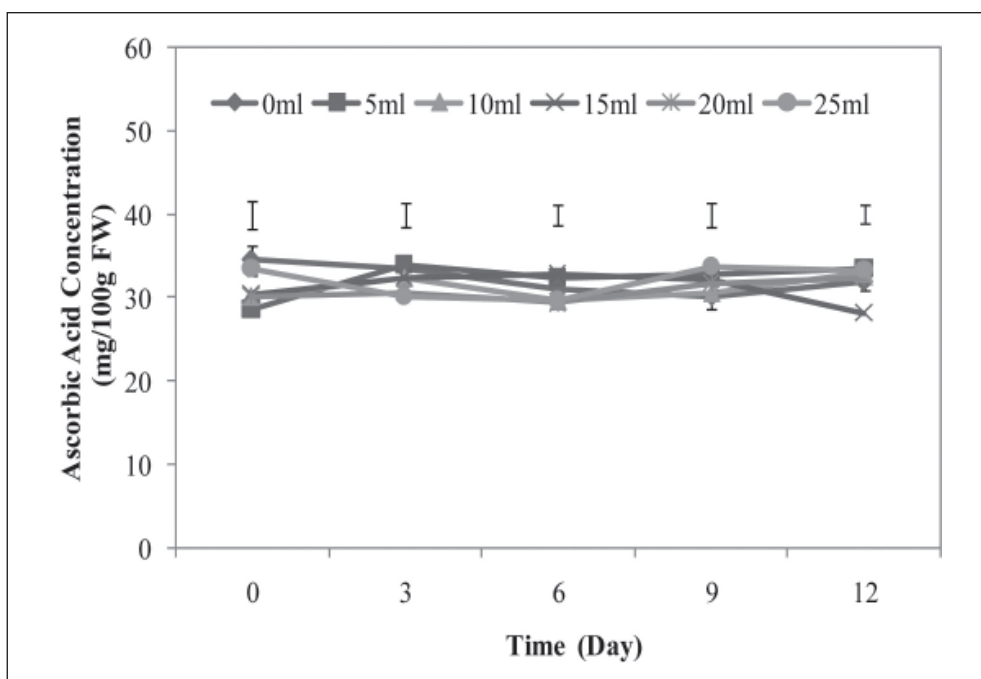


Fig. 8. Effect of different volumes of nitrogen gas fumigation on ascorbic acid concentration of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value on Day 0=3.49, Day 3=2.77, Day 6=2.37, Day 9=2.87 and Day 12=2.19).

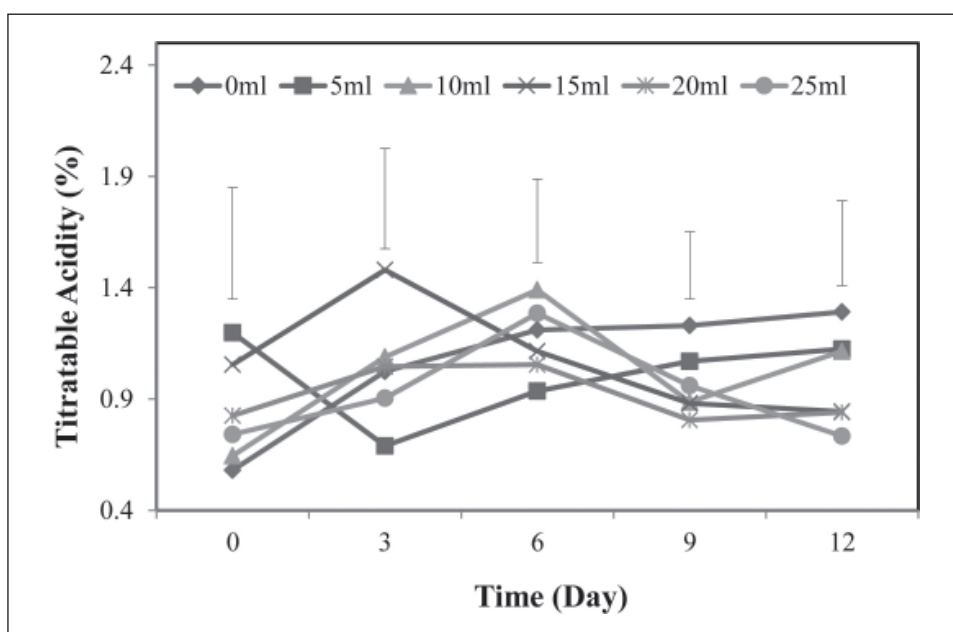


Fig. 9. Effect of different volumes of nitrogen gas fumigation on soluble solids content of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=0.81, Day 3=1.02, Day 6=0.61, Day 9=0.99 and Day 12=0.69)

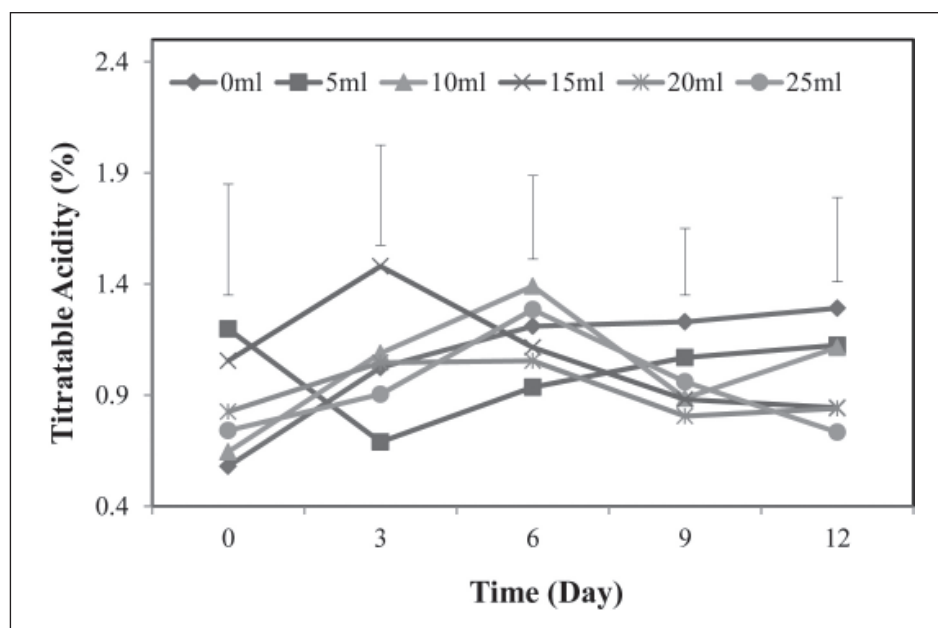


Fig. 10. Effect of different volumes of nitrogen gas fumigation on titratable acidity of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=0.50, Day 3=0.45, Day 6=0.38, Day 9=0.30 and Day 12=0.38)

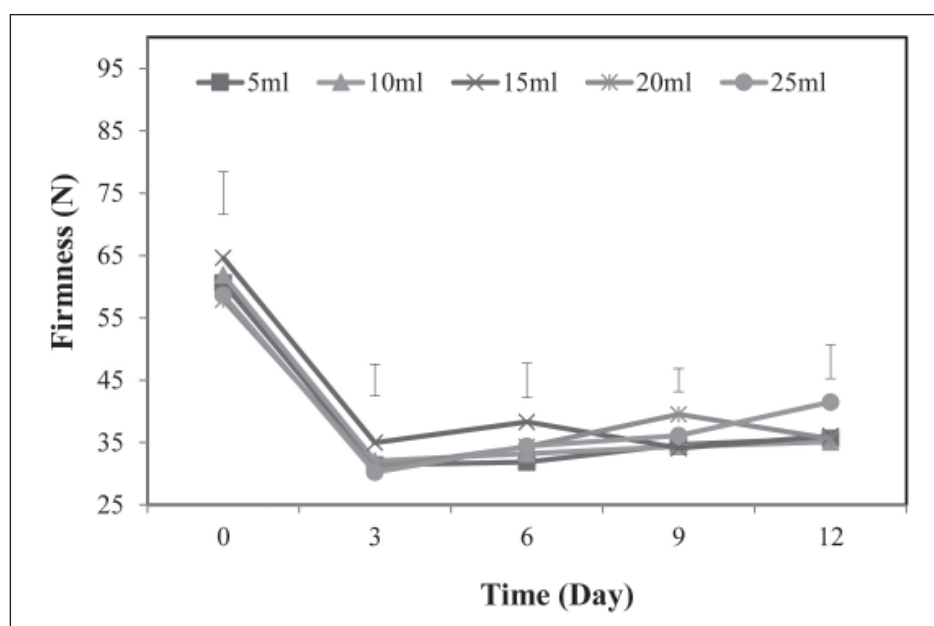


Fig. 11. Effect of different volumes of nitrogen gas fumigation on firmness of MP starfruit. Vertical bars represent LSD at $P \leq 0.05$. (LSD value Day 0=16.82, Day 3=5.11, Day 6=5.50, Day 9=3.71 and Day 12=5.48).

The flesh firmness of MP starfruit fumigated with N_2 gas was declined sharply after 3 days storage in all treatments but tend to remain stable from day 3 to day 12 (Figure 11). The greatly declined in flesh firmness on treated MP starfruits on day 3 may be due to the moisture loss, reduces endogenous protection from the loss of turgor, the increase loss of cell sap or higher transpiration that induced by

the cutting (Ding *et al.*, 2007). According to Rocha and Morais (2003), the firmness of MP apple decreased 50% after 7 days storage at 4°C. These results showed that there is no specific trend can be deduced on flesh firmness of MP starfruits throughout the 12 days experiment and further stored at 5°C. Similarly, Wu *et al* (2012a) found that there are no differences in flesh firmness of pineapple

wedges throughout 20 days at 4°C when treated with inert gas such as high pressure N₂ gas or normal atmospheric-pressure N₂ gas.

CONCLUSION

No adverse effect of N₂ gas was observed on the physical and chemical characteristics of MP starfruit stored at 5°C but it reduces the occurrence of browning incidence. The N₂ gas (25 mL) had a tendency to effectively reduce the browning incidence and maintained other quality attributes of MP starfruit. In addition, the shelf life of N₂ fumigated the MP starfruit also can extend up to 12 days of storage without significant reduction ascorbic acid concentration.

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